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***FINAL REPORT***

**Biological Control of Leafy Spurge, *Euphorbia esula* L :  
Impacts of Eight Rangeland Grasshopper Insecticide Treatments  
on *Aphthona lacertosa* (Rosh.) and *A. nigriscutis* Foudras  
(Coleoptera:Chrysomelidae) in Western North Dakota**

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## Abstract

Established populations of introduced *Aphthona spp.* on leafy spurge may be in jeopardy on western rangelands where populations of grasshoppers require insecticide treatments. Laboratory bioassays and field evaluations were conducted to determine the impacts of grasshopper control treatments. In laboratory bioassays, diflubenzuron spray produced no significant mortality. Malathion and carbaryl sprays produced 17%-67% and 80%-96% significant mortality respectively. In the season of treatment, combined field evaluations showed carbaryl bait, diflubenzuron, malathion and carbaryl sprays resulted in 17%, 0%-18%, 21%-24% and 60%-82% adjusted percentage mortality respectively. *Aphthona spp.* populations in the following year did not decline and year one population decreases did not translate into plant density increases a year after treatment. *Aphthona spp.* field populations exceeded first year pretreatment levels, in 23 of 24 plots one year after treatment. When locations were combined, all treatments except malathion 8 ozs and diflubenzuron 1 oz, resulted in population increases greater than in untreated plots. Reduced treatments of diflubenzuron and malathion resulted in greater population growth at one year after treatment compared to the traditional doses. *Aphthona spp.* populations increased the most in bran bait plots (4.50X), followed by carbaryl 16 oz plots (4.32X), malathion 4 oz plots (3.28X), carbaryl 8 oz plots (2.83X), diflubenzuron 0.75 oz plots (2.73X), untreated check plots (2.08X), malathion 8 oz plots (2.01X) and diflubenzuron 1 oz plots (1.84X). Timing of grasshopper treatments at third instar and peak adult *Aphthona spp.*, allowed for pretreatment oviposition sufficient to insure the survival of the next years generation of *Aphthona spp.*

## Introduction

A native to Europe and Asia, leafy spurge, *Euphorbia esula* L. is an aggressive, perennial weed infesting millions of acres of western rangelands in the US. It easily out-competes native vegetation often destroying diversified plant communities essential for wildlife and generally renders infested rangeland unusable for grazing livestock because of the irritating chemicals produced by the plant.

Introduced into the US in Massachusetts in or before 1827, it had spread to all of the Canadian provinces except Newfoundland by 1950 and to 30 US states by 1979. Infestations can double in acreage every 10 years with the greatest density and damage occurring in the northern Great Plains. Infested rangeland acreages in North Dakota, South Dakota, Montana, and Wyoming were estimated to be 1,624,500 acres by 1994 with the annual indirect and direct impacts of this plant estimated at \$129.5 million for these states alone (Leitch, et al. 1994)

Chemical control is impractical because economic returns from rangeland are negated by the cost of herbicides and application fees over large acreages. However, the recently released natural biological control agents (*Aphthona* flea beetles) from Yugoslavia and Hungary, in North Dakota have demonstrated impressive and widely accepted success. While the first exotic flea beetle released was *Aphthona flava* in 1985, this species was soon replaced by the more easily established species, *A. nigriscutis* in 1989 and *A. lacertosa* in 1993. Although agricultural economists predicted that leafy spurge would continue to expand its range in ND, SD, MT and WY until ca. 2000, the biological control methodologies utilizing these species are predicted to reduce the leafy spurge infestations by 65% (1.21 million acres) on wildlands and rangelands combined by 2025 (Bangsund et al. 1999).

The western rangelands where leafy spurge occurs also support damaging populations of grasshoppers. Grasshoppers are the principal invertebrate consumer of forage on 665 million acres of rangeland in the western U.S. (Hewitt et al. 1976). While these infestations of grasshoppers tend to be cyclic, they may be widespread, covering thousands to millions of acres, that may require chemical intervention. From 1975 thru 2000 alone, the federal government has been involved in cooperative (private, state and federal) control efforts on 36,047,584 acres, ranging from 3,418 acres to 13,687,585 acres per year and averaging 1,386,445 acres per year (USDA, 2002). Private landowners have added additional substantial acreages of rangeland treated each year to these totals.

Because both pests may occur in the same areas, biological control agents established for leafy spurge appear to be at risk when treatments to prevent grasshopper damage are required. These concerns have only recently surfaced with the successes of the introduced *Aphthona* beetles. The primary questions of concern are: Do treatments applied for controlling grasshoppers on rangeland infested with leafy spurge cause mortality to established adult flea beetle biological control agents? Which treatments, if any, do not cause mortality? Of those that do, what is the immediate mortality level to biological control agents that may be exposed? What is the resultant level of suppression on the population of biological control agents at one year after treatment? And, how long is required for the affected population to return to pretreatment population levels?

The following laboratory and field studies were conducted to provide answers to these concerns and to allow land managers to optimize investments in controlling both pests on western rangelands.

## Methods and Materials

### *Treatments.*

The treatments studied were those approved for use in USDA, APHIS sponsored cooperative (private, state and federal) grasshopper and Mormon cricket control programs, termed traditional treatments or reduced agent area treatments (RAATS). Traditional treatments rely on uniform total coverage treatment of the infested area with a goal of maximum mortality to the pest (Foster 1996-1999; Foster et al. 2001). RAATS treatments rely on substantially reducing the dose and leaving alternating areas not directly treated between each treated swath and has a resulting positive cost benefit ratio as a goal (Larsen and Foster, 1996-1999; Lockwood et al. 2000; Foster et al. 2001 ). The alternating not directly treated area not only reduces cost but has the potential to conserve non-targets (USDA, 2002).

The traditional treatments studied were: (1) malathion (Fyfanon ULV) at 8 fluid ozs/acre, (2) carbaryl (Sevin XLR Plus) at 16 fluid ozs plus 16 ozs of water/acre, (3) diflubenzuron (Dimilin 2L) at one fluid oz/ acre plus 10 ozs of Clean Crop Oil Concentrate and 20 ozs of water/acre and (4) 2% carbaryl bran bait (Eco Bran) at 8 lbs/acre (lab) and 2 lbs/acre (field). The RAATS treatments studied were: (1) malathion (Fyfanon ULV) at 4 fluid ozs/acre applied to 80% of the acreage in alternating treated swaths and untreated areas. Treating 80% of the of the acreage was achieved by calibrating the aircraft for a 100 feet wide swath and spacing the aircraft at 125 feet during treatment application. (2) carbaryl (Sevin XLR Plus) at 8 fluid ozs plus 8ozs of water/acre applied to 50% of the acreage. Treating 50% of the acreage was achieved by calibrating the aircraft for 100 feet wide swath and spacing the aircraft at 200 feet during treatment application. (3) diflubenzuron (Dimilin 2L) at 0.75 fluid ozs/acre plus 7.5 fluid ozs of Clean Crop Oil Concentrate and 15 ozs of water/acre to 50% of the acreage. In both laboratory and fields studies, RAATS treatments consisted of the amount of material that would be applied directly below the aircraft. This component of the treatment presents the worst case scenario in terms of potential exposure and impact. In laboratory studies, both malathion treatments were diluted with canola oil and sprayed in a total volume of 12 fluid oz/acre to facilitate the spraying of these lowest total volume/acre treatments. All other liquid laboratory sprayed treatments were identical in diluent ratios to field applied treatments.

### *Laboratory Studies*

Laboratory studies were used to evaluate the mortality produced by treatments when sprayed directly on the adult insects or on vegetation that subsequently hosted unsprayed insects. All 7 treatments were evaluated against *A. lacertosa* and two treatments (carbaryl bran bait and RAATS malathion 4 oz) were evaluated against *A. nigriscutis*. The studies were conducted in the USDA-APHIS-PPQ Bismarck, North Dakota laboratory from June 19, 2000 through June 28, 2000.

*Design.* The experimental design consisted of 21 treatments each involving 10 *Aphthona* adults and each replicated 20 times. The treatments were *A. lacertosa* sprayed with each of the treatments and caged on untreated leafy spurge; Untreated *A. lacertosa* caged on vegetation treated with each of the treatments; *A. lacertosa* caged on untreated vegetation and bait; Untreated *A. lacertosa* caged on untreated vegetation as controls; *A. nigriscutis* treated with carbaryl bran bait and RAATS malathion and caged on untreated leafy spurge; untreated *A. nigriscutis* caged on treated vegetation; and untreated *A. nigriscutis* caged on untreated vegetation as controls.

*Procedures.* Both species were field collected from the Bryan Durham ranch near Sentinel Butte and transported to the laboratory where they were stored in a refrigerator (37 to 41 degree F.). Insects were then sorted to species in the laboratory, placed in groups of 10 in ventilated 4 oz specimen cups containing a fresh cutting of leafy spurge and replaced in the refrigerator. Groups of ten insects at a time were transferred to and treated in a separate room modified to accommodate a spray tower and associated activities. Insects were placed on a paper covered chill plate to facilitate handling during treatment. A clean paper was used for each separate spray application. The spraying system (**Fig. 1**) consisted of a tower mounted air brush (Paasche Type H with R 75 regulator) modified with customized siring needles for liquid injection to produce droplets which simulate aerially applied treatments (Foster and Reuter 1991; Foster et al. 1996-1999).

Individually treated young leafy spurge plants which were to receive untreated insects were similarly handled except that no chill plate was used. Plants were 30 to 40 cm high and propagated from root cuttings in Container, tree propagation pots (6.3 cm ID x 25.4 cm high). Untreated insects included in the experimental design were similarly exposed to the chill plate in the study. Insects and individual leafy spurge plants were then placed in cages and maintained in the laboratory. Cages (6.3 cm OD x 45.72 cm high) were constructed from 6.3 cm OD clear PVC pipe screened on one end with organdy. Cages were secured to the propagation pots with duct tape and were monitored daily for mortality for 7 days.

During the study maximum daily temperatures ranged from 66.3 to 75.9 and averaged 71.0 degrees F. Minimum daily temperatures ranged from 62.2 to 67.7 and averaged 64.6 degrees F. Maximum daily humidity ranged from 67.3 to 83.8 and averaged 76.6 percent. Minimum daily humidity ranged from 34.9 to 60.6 and averaged 47.7 percent.

### *Field Study*

Grasshopper control treatments operationally applied to small plots of leafy spurge that had been established within the last three years with *Aphthona spp.* were evaluated over the 2000 and 2001 summer seasons.

*Study area.* Three separate locations in the Little Missouri River drainage of western North Dakota were used for the study (**Fig.2**). The center of the northern most location was 5.5 miles east and 4 miles north of Sentinel Butte on the Bryan Durham ranch in Golden Valley County. The center of the most central location was 17 miles west and 10 miles north of Amidon on the Gary Van Daele ranch in Golden Valley and Slope Counties. The center of the southern most

location was 6.5 miles west and 2 miles north of Amidon on the Wilber Aus ranch in Slope county. The most central location was 14 miles from the southern location and 24.5 miles from the northern location. The general locations were selected because of the history of leafy spurge and grasshoppers and the recent establishment of *Aphthona* beetles on small stands of leafy spurge.

Plots at the Durham location were categorized as belonging to the Cabbart – Cherry association (shallow and deep well drained, medium textured, gently sloping to very steep soils) and the Badland – Cabbart association (badland and shallow and deep, well drained, medium textured, gently sloping to very steep soils). Plots at the Van Daele location were categorized as belonging to the Hadley Glendive association (deep, somewhat excessively drained and well drained, moderately coarse textured, level and nearly level soils) and Badland – Cabbart association (badland and shallow and deep, well drained, medium textured, gently sloping to very steep soils). Plots at the Aus location were categorized as belonging to the Brandenburg – Cabba – Cabbart association (well drained to excessively drained, shallow soils that are medium textured) (Thompson et al. 1978; Aziz et al. 1989). Soil types of each plot at each of the locations is shown in **Table 1**.

*Design.* At each location, eight 0.23 acre (100 ft x100 ft) plots of leafy spurge stands containing mixed populations of *A. nigriscutis* and *A. lacertosa* were established to accommodate each of the treatments and an untreated control (**Figs. 3-5**). Plots were separated from adjacent plots by at least 200 yards and from adjacent stands of leafy spurge by at least 50 yards. A weather station was established at each of the three locations to record precipitation, humidity, and min and max temperatures for each day from July 2 – July 30, for the season of treatment.

*Treatment applications.* All liquid field treatments were applied with an USDA, APHIS aircraft (Cessna Ag-Truck equipped with a standard commercial spraying system) and was operated by an APHIS pilot who was highly experienced with precision work for research. The aircraft was also equipped with differentially corrected guidance and recording systems. However, primary guidance was provided by ground personnel that measured each swath and gathered meteorological data during application. The aircraft was additionally equipped with winglets (DBA-Ag Tips; Clark Oberholtzer, Alberta Canada). With a swath width and plot width of 100 feet, complete coverage of the plot was insured by flying two passes on each plot. One pass down each of two opposite plot boundaries for a short distance beyond the corner. While the plot size was 0.23 acre, the actual area treated was ca. one acre for each plot (**Fig. 6**). Oil or water sensitive spray cards were placed ca. every 17 feet along the plot boundaries situated perpendicular to the flight line to insure complete coverage had occurred during application. Prior to application, the aircraft spray system was calibrated to operate under parameters which resulted in delivery of spray within one percent of the desired rate per acre for each of the treatments applied. Calibration for each of the treatments was accomplished by collecting and measuring the amount of material sprayed through each nozzle for each treatment set up, for a predetermined amount of time and making adjustments in pressure until the desired output was achieved and replicating this procedure three times.

Liquid treatments were applied through flat fan Tee Jet stainless steel nozzle tips directed straight down. The traditional and RAATS malathion treatments were applied at 120 mph and 46

psi with 8 and 4 (8002) size tips respectively on July 9, 2000. The traditional and RAATS carbaryl treatments were applied at 120 mph and 41 psi with 20 and 10 (8003) size tips respectively on July 7, 2000. The traditional and RAATS diflubenzuron treatments were applied at 125 mph and 36 psi with 20 and 15 (8003) size tips respectively on July 5, 2000. All treatments were applied from an altitude of 30 to 50 feet. Winds during application ranged from < 1 to 4 mph and averaged < 2 mph for all plots combined. Other plot specific meteorological conditions recorded during application are summarized in **Table 2**. Daily precipitation records for the month of July for all three locations are summarized in **Figure 7**. The carbaryl wheat bran bait treatment was ground applied with a Scotts Handy Green II hand spreader modified with a hopper cover. Sawdust at 2:1 ratio (sawdust:bait) was used as a diluent to facilitate calibration and a uniform application. The applicator was calibrated for a 5 feet swath width applied at a walking speed of ca. 5 mph which was practiced extensively before actual field application. This accommodated 20 passes per plot. Guidance was provided by ground personnel. The hopper accommodated material for ca. 3.5 swaths. Each bait plot required 20 swaths.

*Aphthona population estimates.* Combined densities of adult *A. lacertosa* and *A. nigriscutis* were estimated at each of 16 fixed and uniformly distributed sampling sites within each of the plots (**Fig. 6**). Each site was marked with a numbered stake to identify the specific location within the plot. A standard 15 inch dia. sweep net was used to take two 180 degree sweeps at each site. The number of *A. lacertosa* and *A. nigriscutis* combined were counted from the sweep net bag at each stake immediately upon completion of both sweeps. Upon completion of the count and without moving from the stake, all captured insects were released at that site before moving forward to the next sampling site within the plot. The same individual conducted all sweeps and counts for all sites and plots for the duration of the study. Two pretreatment and three posttreatment counts were conducted during the initial year (2000) of the study at weekly intervals from June 23 thru July 26. Nine weekly post counts were conducted from June 4 thru Aug. 1 during the second year (2001) of the study. All of the treated plots and the untreated plot (total 8 plots) at each location were counted on the same day. A sweep sample (consisting of 2-4 sweeps to minimize impact on total plot population levels) was taken at the center of each site between June 29 thru July 7, 2000, around peak adult presence and returned to the laboratory for identification to determine *Aphthona spp.* composition.

*Grasshopper species and age structure.* The abundance of each grasshopper species and the associated age structure was determined at each of the three locations from sweep samples taken near the center of each location on the day of treatment. Each sample consisted of 100 low and slow sweeps and 100 high and fast sweeps combined (Foster and Reuter 1996.) Low and slow sweeps performed at ground level insured capture of very young instar stages and less active grasshopper species while high and fast sweeps performed at the canopy of the vegetation insured capture of older instar stages and more active species. After collection, samples were cold stored until they could be sorted and identified in the laboratory. Mean grasshopper developmental age was calculated by multiplying the number of grasshoppers in each succeeding stage, instars 1 - 5 and adult, by values of 1 – 6 respectively and dividing that value by the total number of grasshoppers in all stages multiplied by 6 to arrive at a maturity index. This value was then multiplied by 6 to arrive at the mean developmental age. (Mean population age =  $(1(a) + 2(b) + 3(c) + 4(d) + 5(e) + 6(f)/6(a+b+c+d+e+f+g)) \times 6$ , where a-f equal the number of grasshoppers in each succeeding stage, instars 1-5 and adults.

*Leafy spurge density index.* A density index was developed to describe the leafy spurge stand at each of the 16 sampling sites for all plots and locations. Values of 1 to 4, determined visually at each sweep location, were assigned to each of the sites before and ca. one year after treatments. A value of one was assigned to a site with no leafy spurge plants in the sweep area; two to a site with one to a few plants in the sweep area; three to a site with many plants but not continuous coverage in the area; and four to a site with continuous plants in the sweep area (**Fig. 8**). Density indexes were generated pre treatment on July 2 - July 7, 2000 and post treatment on June 20 – 25, 2001 and July 30, – Aug. 1, 2001.

### *Data Analysis*

Laboratory studies relied on one way analysis of variance of ranked percentage mortality values with a Tukey multiple comparison procedure to separate differences.

Population data of *Aphthona spp.* was analyzed separately for each field location. Ranks of percentage mortality values and percentage control values of *Aphthona spp.* in the year of treatment and ranks of fold increases in *Aphthona spp.* populations between years were analyzed with a one way analysis of variance and Tukey multiple comparison procedure.

Percentage control data was developed by adjusting population reductions in treated plots with the natural mortality measured in untreated populations (Connin and Kuitert, 1952). Adjusted percentage control of the treatment (which takes into account natural changes in the untreated population) was calculated by the formula  $100 (1 - Ta \times Cb/Tb \times Ca)$ . Where Tb equals the total population of adult *Aphthona* counted before treatment, Ta equals the total counted after treatment, Cb equals the total counted for the untreated control sites before treatment, and Ca equals the total counted for the untreated sites after treatment.

Plant density index data were expressed as percentage or fold increases, and were evaluated by either a Kruskal-Wallis with Dunn's method (an all pair wise multiple comparison procedure) or an one way analysis of variance with a Tukey multiple comparison procedure used to determine differences between treatments (SPSS Inc. 1977).

## Results and Discussion

### *Laboratory Studies-2000*

*Treated insects.* When insects were treated directly and placed on untreated vegetation, all treatments at 3 - 7 days after application except diflubenzuron, produced mortality statistically higher than occurred in the untreated controls (**Table 3**). All carbaryl spray and bait treatments resulted in mortality statistically higher than malathion treatments. Carbaryl spray produced mortality ranging from 91% - 95% while bait produced mortality ranging from 67% - 90%. However, Carbaryl spray and bait treatments were not statistically different for *A. lacertosa* but were significantly different for *A. nigriscutis*. Malathion treatments produced mortality ranging from 25% - 41%. Diflubenzuron produced mortality ranging from 6% - 27%, numerically but not statistically higher than untreated controls. Significant mortality to adults was not expected with diflubenzuron, an insect growth regulator that inhibits the production of chitin and causes



mortality while molting from one developmental stage to the next. The high mortality produced by carbaryl bait was somewhat surprising and indicates substantial ground feeding activity by the beetles can occur since all bait was found on the substrate of the cages. However, other ground dwelling beetles have been shown to be susceptible to carbaryl bran bait (Quinn et. al 1990; Quinn et. al 1991). Mortality in untreated controls was low, ranging from 1% – 1.4%/day.

The higher traditional dose of each insecticide produced mortality numerically higher than the lower RAATS dose in all cases, except carbaryl spray at 4, 6 and 7 days after treatment. However, those results were essentially identical. Treatments common to *A. nigriscutis* and *A. lacertosa* resulted in statistically similar mortality, apparently indicating equivalent species susceptibility. All treatments and the untreated controls demonstrated slightly increasing mortality from 3 – 7 days after treatment. However, increases with malathion and diflubenzuron treatments were not commensurate with untreated increases.

*Treated vegetation.* Generally, these results were very similar to those of treated insects. When untreated insects were placed on treated vegetation, all treatments at 3 - 7 days after application except diflubenzuron and the low dose of malathion presented to *A. lacertosa* produced mortality statistically higher than occurred in the untreated controls (**Table 4**). Carbaryl spray produced mortality ranging from 80% - 96% and both carbaryl treatments resulted in statistically higher mortality than any malathion treatment. Malathion treatments produced mortality ranging from 17% - 67%. and were numerically and usually statistically higher than untreated and diflubenzuron treated populations, except for *A. lacertosa* exposed to the 4 oz malathion vegetation treatment. Diflubenzuron produced mortality ranging from 8% – 26%, numerically but not statistically higher than the untreated controls. It is important to note that beetles were unexpectedly found more frequently on the cage substrate of diflubenzuron treated vegetation compared to other treatments and controls. Mortality in untreated controls was low, ranging from 1% – 1.3%/day.

In all cases, each insecticide performed in dose rank order. Unexpectedly, the malathion 4 oz treatment resulted in significantly higher mortality to *A. nigriscutis* (63% - 67%) compared to *A. lacertosa* (17% - 25%). This was dissimilar to results seen when insects only were treated. However, it was noted in this study that *A. nigriscutis* positioned themselves higher on the plant in the cages compared to *A. lacertosa*. Since the upper most part of each plant could receive more insecticide during application, positioning in the uppermost area of the plants could lead to greater exposure explaining the higher mortality. This seems reasonable since no difference in resulting mortality was detected between species when they were sprayed directly and there was no difference in mortality between species in untreated populations. Again, all treatments and the untreated controls demonstrated slightly increasing mortality from 3 – 7 days after treatment with diflubenzuron treatment increases not commensurate with untreated increases.

#### *Field Studies- 2000*

*Adult Aphthona populations.* Although the population densities were different at the three locations, changes that occurred over the season of treatment in untreated control populations at each of the three locations were similar (**Fig. 9**). Adult populations peaked in the Aus, Van Daele and Durham locations on July 2, July 5 and July 12 respectively. All treatments had been applied

within a week of peak adult *Apthona spp.* At the Aus location, treatments occurred from 3 to 7 days after the adult population peaked. At the Van Daele location treatments occurred from 0 to 4 days after the adult population peaked. At the Durham location treatments occurred from 3 to 7 days before the adult population peaked.

The rapid natural decline in untreated populations at two and three weeks after treatment (**Fig. 9; Table 5**) confounded and obscured any statistical differences between treatments at those post treatment intervals. However, adult populations closer to peak and pretreatment levels at one week after treatment were more suitable for analyses.

At the Aus location at one week after treatment only carbaryl spray treatments resulted in significantly higher mortality than occurred in the untreated population (**Table 6**). The carbaryl bait treatment resulted in significantly less mortality than carbaryl or malathion sprays. All treatments performed numerically in dose rank order at this location.

At the Van Daele location at one week after treatment no treatments resulted in mortality significantly different than demonstrated in the untreated population and there was no significant difference between any of the treatments (**Table 6**). Again, treatments performed numerically in dose rank order except for the carbaryl 8 oz treatment.

At the Durham location at one week after treatment the high dose of each insecticide spray and the carbaryl bait resulted in significantly higher mortality than occurred in the untreated population (**Table 6**). Low doses of all sprays produced significantly lower mortality than the high doses. However, low dose spray mortalities were not significantly different than mortality in the untreated population.

Populations in some plots increased after treatments occurred (indicated by a negative percentage reduction). Diflubenzuron treated populations increased in 4 of 6 cases while untreated populations increased in 3 of 9 cases.

The diflubenzuron treatments were not expected to cause mortality to adults. Diflubenzuron is an insect growth regulator that interferes with the formation of chitin and as such can cause death to immature forms of insects when they molt. Adults would not be susceptible to death as a result of exposure to diflubenzuron. However, immature individuals or perhaps eggs resulting from adults exposed to diflubenzuron may be at risk. Mortality associated with these stages of the insect would not be evident until the following year. It is important to note that in the case of the one oz dose of diflubenzuron at the Durham location, mortality was higher than in the untreated population (**Table 6**). This occurrence was consistent for the high dose for all three weeks of sampling at the Durham location (**Table 5**). Laboratory observations indicated more adults positioning themselves on the substrate with diflubenzuron treatments compared to other treatments including untreated populations. If any of this behavior occurred in the field, sweep sampling at canopy height would underestimate adult populations because fewer would be in the actual sampling area and thus may explain the reduced population.

The carbaryl sprays that produced significant mortality averaged 73.7 % mortality. Malathion spray, Dimilin spray and carbaryl bait produced significant mortality in only one of the locations, 58%, 28% and 51% respectively.

While results from the three locations varied, when mean percentage mortality values of all locations were combined (**Table 7**) and adjusted for natural mortality at one week after treatments (Connin and Kuitert, 1952), carbaryl spray, malathion, carbaryl bait and diflubenzuron produced high (60-82%), moderate (21-44%), light (17%) and light if any (-36-18%) mortality respectively. These combined results were similar to those seen in the laboratory studies except for carbaryl bait. Field treatments with bait were one fourth the amount applied in laboratory studies, thus explaining reduced field mortality.

While substantial rainfall occurred during the study (**Figure 7**) only the malathion treatments were subjected to rain within a few hours of application. Other experiences by the authors with rain and diflubenzuron or carbaryl treatments have shown little effect on resulting grasshopper control. However, substantial rainfall following a malathion treatment on rangeland can significantly reduce the control efficacy (Foster et al. 1981). Even though rain occurred within a few hours of the malathion treatments, except for the carbaryl bait treatments, these field results were generally consistent with those in the laboratory studies and indicate little effect from the rain on treated leafy spurge plants. (**Table 7**). In the laboratory study, bait treatments produced higher mortality than in the field but occurred at 4 times the field rate.

As expected, some population reduction occurred in the season of treatment. However, substantial oviposition had occurred before treatments were applied. Any substantial impacts on established populations would be most evident in the succeeding generation in the year following treatment and would most likely be a result of a decreased number of eggs and larvae due to reduced parental stocks of adults for reproduction and oviposition. The risk of direct exposure of eggs and resulting larvae to any treatments would have been minimal if at all because of the life cycle of *A. lacertosa* and *A. nigriscutis*. These species are univoltine. In North Dakota, over a period of about 2 months (mid June - mid August) females lay eggs in small batches (50 -200 eggs/female/lifetime) underground near the stem of leafy spurge or on the stem near the surface where they hatch in 10 – 14 days and then seek out young leafy spurge roots to feed upon. The larvae feed and develop through three larval stages, gradually moving to larger roots and buds. With approaching cool weather, third instar larvae move deeper in the soil, where they cease feeding and over winter in a state of diapause until it is broken by warmer soil temperatures in April or May (North Dakota Biological Control Committee, 1998)

*Aphthona species composition.* The Van Daele location showed a greater prevalence of *Aphthona nigriscutis* while the Aus location showed a prevalence toward *A. lacertosa*. The prevalence of both species were similar at the Durham location. When all plots and locations were combined the prevalence of both species were similar (**Table 8**). Because of the small plot size and limited numbers of adults, no attempt was made to differentiate mortality between species in the field which would have necessitated additional sampling and could have impacted the population totals.

*Leafy spurge density 2000.* Mean individual plot density indices ranged from a low of 1.8 at the Aus diflubenzuron 0.75 oz plot to a high of 3.2 at the Durham malathion 8 oz plot (**Table 9**). The average indexes of Aus, Van Daele and Durham plots were 2.5, 2.8 and 2.9 respectively. Prior to treatment there were no significant differences between plant density indices in the plots at the Van Daele or Durham locations. At the Aus location all plots were statistically similar in density indices except for the diflubenzuron 0.75 oz and untreated plots.

*Grasshopper species composition and age structure.* As expected, the assemblage of grasshopper species at each location was different (**Table 10**). Twelve species were common to all locations and of the 5 most abundant species at each location only *Ageneotettix deorum* was common to all 3 locations.. At the Durham location the 5 most abundant species were *Melanoplus packardii* (23%), *A. deorum* (20%), *M. sanguinipes* (17%), *M. femurrubrum* (7%) and *Phoetaliotes nebrascensis* (5%) The mean instar age for all 24 species found at this location at the time of treatment was 3.70 (between the third and fourth instar stages). At the Van Daele location the 5 most abundant species were *P. nebrascensis* (55%), *Opeia obscura* (10%), *M. dawsonii* (8%), *A. deorum* (5%), and *Encoptolophus costalis* (5%). The mean instar age for all 18 species at this location at the time of treatment was 2.52 (between the second and third instar stages). At the Aus location the 5 most abundant species were *P. nebrascensis* (27%), *A. deorum* (12%), *Orphulella speciosa* (9%), *Opeia obscura* (9%), and *M. femurrubrum* (8%). The mean instar age for all 23 species at this location at the time of treatment was 2.97 (the third instar stage).

Developmental phenology of both grasshoppers and *Aphthona spp.* are temperature dependent. Consequently, one would expect locations with the most developed (oldest) grasshoppers to be consistent with the earliest peaking adult *Aphthona* locations. However, because different grasshopper species occur and develop at different times, mean ages of dissimilar assemblages of grasshopper species can be misleading in estimating the rank order of developing adult *Aphthona* populations. A better estimate for predicting order of adult *Aphthona* population development in different locations should rely on estimating assemblage age of grasshopper species common to all locations. The two species common to all three locations and composing at least 5% of each population showed standardized grasshopper assemblage ages developing first at the Van Daele location followed by those at the Durham and Aus locations (**Table 11**). However, these calculations indicate only about 1-2 days difference in ages between locations for common species. The rates of development calculated for grasshoppers is somewhat consistent with the peak adult occurrence of *Aphthona spp.* at the three locations. The peak *Aphthona* populations seemed to occur first in 2000 at the Aus location. However, only minor reductions in the early numbers at this location could have easily placed the Aus location in a similar order as calculated for grasshopper development (**Figure 9**).

The ages of these grasshopper assemblages at all three locations are younger than or near the youngest ages that would be treated in large scale cooperative programs. Generally, control programs occur significantly after the third instar age and may include even a few adults but are conducted before egg deposition commences. Historically in western North Dakota, when seasonal weather conditions have been similar to those in this study, cooperative control programs have occurred during the same time period as in this study, early July.

## Field Studies- 2001

*Aphthona* spp. Adult populations peaked first in the Durham location followed by the Aus and Van Daele, an order dissimilar to that in 2000 (**Figs. 9 & 10**). *Aphthona* spp. populations numerically exceeded the first year (2000) pretreatment and peak adult levels, in 23 of 24 plots one year after treatment (**Figs. 11, 12 and 13**). The peak population in the 8 oz malathion treated plot at the Van Daele location declined 23% from 2000 to 2001 as the exception. Population increases in six of 21 treated plots were significantly greater than population increases in untreated plots (**Table 12**).

At the Aus location, peak 2000 adult populations in plots treated with carbaryl 16 oz/acre, carbaryl bran bait and malathion 4 oz/acre increased significantly more in 2001 than untreated populations. During the same period, the traditional dose (16 oz/acre) of carbaryl resulted in populations increasing significantly more than populations treated with the RAATs dose (8 oz/acre). However, populations treated with the RAATs dose of diflubenzuron (0.75%/acre) increased significantly more than populations treated with the traditional dose (1 oz/acre).

At the Van Daele location only populations treated with carbaryl bran bait increased significantly more than untreated populations from 2000 to 2001. No significant differences in population increases were detected between the RAATs and the parental traditional treatment.

At the Durham location populations treated with both doses of carbaryl spray increased similarly and were significantly greater than untreated populations. The RAATs doses of the malathion and diflubenzuron sprays resulted in population increases from 2000 to 2001, significantly greater than occurred in populations treated with the parental traditional doses.

Intuitively one would expect *Aphthona* populations in treated plots to grow slower than in untreated plots. However, the frequency of exceptions (6 separate cases showed significant increases in treated populations compared to untreated populations and the trend of 13 of 21 plots with populations numerically increasing more than untreated populations), is suggestive of treatments effecting the naturally occurring predators of the introduced *Aphthona* spp. (**Table 12**). However, when considering cases of significant increases and cases of trend increases combined, only the lowest doses of malathion and carbaryl sprays were consistent for all three locations in producing populations increasing more than populations increased in untreated plots. If such is the case, one could consider treating established populations to promote greater speed in establishing populations of *Aphthona* spp. on leafy spurge. However, further study under a broader range of conditions should first be conducted.

When all locations were combined, mean *Aphthona* spp. populations increased the most in bran bait plots (4.50X), followed by carbaryl 16 oz plots (4.32X), malathion 4 oz plots (3.28X), carbaryl 8 oz plots (2.83X), diflubenzuron 0.75 oz plots (2.73X), untreated check plots (2.08X), malathion 8 oz plots (2.01X), and diflubenzuron 1 oz plots (1.84X).

Excluding untreated plots, mean plot increase of *Aphthona* populations was highest (3.52X) at the Van Daele location, followed by the Durham location (3.00X), and lowest (2.68X) at the Aus location. Considering grasshopper age at time of treatment, these increases are consistent with

the increasing age of grasshopper species common to all locations. *Ageneotettix deorum* averaged instar ages of 5.0, 4.8 and 4.6 and *Phoetaliotes nebrascensis* averaged instar ages of 2.2, 2.1 and 1.9 at the Van Daele, Durham and Aus locations respectively. Adult untreated *Aphthona spp.* peaks in untreated plots in 2000 occurred earliest at the Aus location, followed by the Van Daele location and the Durham location. However, only minor reductions in the early numbers at this location could have easily placed the Aus location in a similar order as calculated for grasshopper development (**Figure 9**). In 2001 adult untreated populations peaked first at Durham followed by Aus and finally the Van Daele location indicating a difference in order of accumulated heat units occurred in the locations between years.

*Leafy spurge density 2001*. Even though no statistical decreases in *Aphthona* populations occurred in treated plots compared to untreated plots, some numerical reductions were observed (**Table 12**). If reductions in adult *Aphthona spp.* populations in the year of treatment were to have an impact on the next generation, the impact was expected to be expressed as plant density increases one year after treatment. However, percentage increases in plant density indices were not statistically greater than the associated untreated control plot at any time or location (**Table 13**). The indices not only indicate that none of the treatments studied resulted in plant populations increasing faster than in untreated plots but suggest that some treatments may assist in reducing plants quicker compared to untreated plots.

At the Aus location all plots showed similar plant density increases except the carbaryl 8 oz plot which exhibited a significantly lower plant density increase at the near peak adult (early) *Aphthona spp.* interval (**Table 13**). At the Van Daele location, all plots at both evaluation intervals showed similar plant density increases. At the Durham location, all plots showed similar plant density increases except the high dose of Dimilin at both evaluation intervals, high dose of malathion near peak adult (early) *Aphthona spp.* interval and bran bait at the late season evaluation. In all three cases, plant density increases were significantly less than the untreated control.

When all locations were combined, mean end of season numerical increases in plant density indices were highest in the diflubenzuron 0.75 oz and malathion 4 oz plots (+ 9%), followed by the carbaryl 16 oz plots (+ 3%). Untreated plots did not change. Plant indices numerically decreased in carbaryl 8 oz plots, (- 1%), bran bait and malathion 8oz plots (- 12 %), and diflubenzuron 1 oz plots (- 18 %).

## Conclusions

Laboratory studies with 7 different grasshopper control treatments demonstrated *Aphthona spp.* mortality resulting from both direct impingement of spray droplets and ingestion of sprayed vegetation. Field studies in the year of treatment generally demonstrated similar results with the same treatments. However, *Aphthona spp.* populations in the following year did not decline and decreases in year one populations did not translate into increasing plant densities a year after treatments occurred. Field populations in the year after treatment showed remarkable resiliency to all treatments, and in 23 of 24 plots demonstrated a population increase over the previous year.

In North Dakota, timing of grasshopper treatments at about third grasshopper instar and peak adult *Aphthona spp.*, allowed for pretreatment oviposition of sufficient magnitude to insure the survival of the next generation of *Aphthona spp.* in the succeeding year. Grasshopper treatment programs that usually occur later than third instar would pose even less of a threat to established populations of *Aphthona spp.* as greater numbers of eggs would have been oviposited and progeny protected before treatment occurred. These data strongly indicate that these treatments will not cause significant declines in established populations of *Aphthona spp.* and additionally suggest that some of these treatments may promote faster *Aphthona spp.* population increases than in untreated locations. Additional study is warranted to determine if this increase is a result of controlling existing predators of the *Aphthona spp.* This study presents data that will allow the land owner and/or manager to optimize both grasshopper and leafy spurge control efforts to improve overall range and wild land quality.

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Table 1. Soil type descriptions for the three study areas in Golden Valley and Slope counties.

Treatment	Site	Soil type description
<b>Durham</b>		
Untreated	1	24B – Cherry silt loam, 3 – 6% slopes
Dimilin ¾oz	2	21C – Chamma silt loams, 3 – 9% slopes
Dimilin 1oz	3	3 – Harver silt loam, channeled
Carbaryl 16oz	4	10F – Cabbart – Badland complex, 15 – 50% slopes
Carbaryl 8oz	5	24B – Cherry silt loam, 3 – 6% slopes
Malathion 4oz	6	24B – Cherry silt loam, 3 – 6% slopes
Malathion 8oz	7	19F – Cabbart – Cherry silt loams, 9 – 35% slopes
Bran Bait	8	19F – Cabbart – Cherry silt loams, 9 – 35% slopes
<b>Van Daele</b>		
Malathion 8oz	1	HaA – Hanly soils, 1 – 3% slopes
Carbaryl 8oz	2	83F – Badlands – Cherry complex, 6 – 75% slopes
Carbaryl 16oz	3	83F – Badlands – Cherry complex, 6 – 75% slopes
Untreated	4	BaF – Badland Cabbart complex, 9 – 50% slopes
Malathion 4oz	5	HaA – Hanly soils, 1 – 3% slopes
Dimilin 1oz	6	GIA – Glendive fine sandy loam, 1 – 3% slopes
Dimilin ¾oz	7	GIA – Glendive fine sandy loam, 1 – 3% slopes
Bran Bait	8	GIA – Glendive fine sandy loam, 1 – 3% slopes
<b>Aus</b>		
Carbaryl 8oz	1	Kh – Korchea and Harve soils, channeled
Malathion 4oz	2	Kh – Korchea and Harve soils, channeled
Malathion 8oz	3	Peb – Patent loam, 3 – 6% slopes
Carbaryl 16oz	4	Kh – Korchea and Harve soils, channeled
Bran Bait	5	GIA – Glendive fine sandy loam, 1 – 3% slopes
Dimilin 1oz	6	GIA – Glendive fine sandy loam, 1 – 3% slopes
Untreated	7	GIA – Glendive fine sandy loam, 1 – 3% slopes
Dimilin ¾oz	8	GIA – Glendive fine sandy loam, 1 – 3% slopes

Table 2. Meteorological conditions recorded during aerial application of treatments.

Treatment	Date	Plot no.	Time (AM)		Temperature ° F			Wind-mph
			Start	End	Ground	Air	Aircraft	
<b>Aus</b>								
Dimilin 1oz	5-Jul	6	5:30	5:34	50.0	51.0	58.0	2.0 – 4.0
Dimilin 3/4oz	5-Jul	8	6:55	7:00	57.0	68.0	58.0	< 1.0
Carbaryl 16oz	7-Jul	4	5:35	5:40	55.0	59.0	65.0	< 0.5
Carbaryl 8oz	7-Jul	1	6:15	6:18	62.5	63.5	68.0	0.5 – 1.5
Malathion 8oz	9-Jul	3	6:10	6:15	67.0	67.0	65.0	0.5 – 2.0
Malathion 4oz	9-Jul	2	7:35	7:39	73.0	74.0	68.0	2.5 – 3.0
<b>Van Daele</b>								
Dimilin 1oz	5-Jul	6	5:16	5:20	52.0	50.0	54.0	1.0 – 1.5
Dimilin 3/4oz	5-Jul	7	6:15	6:20	52.0	53.5	56.0	< 1.0
Carbaryl 16oz	7-Jul	3	5:25	5:29	58.0	58.0	58.0	1.5 – 2.0
Carbaryl 8oz	7-Jul	2	6:22	6:25	60.0	59.0	63.0	< 1.0
Malathion 8oz	9-Jul	1	5:58	6:02	66.0	66.0	68.0	1.0 – 4.0
Malathion 4oz	9-Jul	5	6:55	7:00	68.0	68.0	63.0	< 1.0
<b>Durham</b>								
Dimilin 1oz	5-Jul	3	4:55	5:00	52.1	52.8	68.0	< 1.0
Dimilin 3/4oz	5-Jul	2	6:30	6:34	54.5	55.1	60.0	< 1.0
Carbaryl 16oz	7-Jul	4	5:10	5:14	60.6	62.2	62.0	< 1.0
Carbaryl 8oz	7-Jul	5	6:36	6:40	62.9	64.4	64.0	4.0
Malathion 8oz	9-Jul	7	5:44	5:46	65.7	66.2	68.0	1.0 – 3.0
Malathion 4oz	9-Jul	6	6:12	6:15	66.4	66.8	66.0	1.0 – 4.0

Table 3. Mortality of *Apthona* flea beetles when treated directly and placed on untreated vegetation in a laboratory study.

Treatment (insects) <sup>1</sup>	Days after treatment – mean % mortality <sup>2</sup>				
	3	4	5	6	7
Carbaryl 16oz – AL	93 a	93 a	94 a	94 a	94 a
Carbaryl 8oz – AL	91 a	94 a	94 a	95 a	95 a
Carbaryl bait – AL	83 ab	84 ab	87 ab	89 ab	90 ab
Carbaryl bait – AN	67 b	70 b	70 b	71 b	71 b
Malathion 8oz – AL	30 c	32 c	38 c	41 c	41 c
Malathion 4oz – AL	25 cd	27 cd	28 cd	30 c	32 cd
Malathion 4oz – AN	32 c	35 c	35 cd	35 c	36 c
Dimilin 1oz – AL	8 de	12 de	17 de	24 cd	27 cde
Dimilin 3/4oz – AL	6 e	7 e	7 e	8 d	12 de
UTC – AL	4 e	6 e	7 e	9 d	9 e
UTC – AN	3 e	4 e	5 e	6 d	7 e

<sup>1</sup> Applied treatment materials to insects only. AL = *Apthona lacertosa*, AN = *Apthona nigriscutis*

<sup>2</sup> A one-way analysis of variance with a Tukey multiple comparison procedure determined statistical differences ( $P \leq 0.05$ ). Means in the same column followed by the same letter are not significantly different.

Table 4. Mortality of untreated *Aphthona* flea beetles placed on treated vegetation in a laboratory study.

Treatment (vegetation) <sup>1</sup>	Days after treatment – mean % mortality <sup>2</sup>				
	3	4	5	6	7
Carbaryl 16oz – AL	89 ab	92 ab	93 a	94 a	96 a
Carbaryl 8oz – AL	80 b	81 b	84 a	85 a	86 ab
Malathion 8oz – AL	29 d	30 d	34 c	36 c	38 c
Malathion 4oz – AL	17 de	18 de	19 cd	23 cd	25 cd
Malathion 4oz – AN	63 c	63 c	64 b	65 b	67 b
Dimilin 1oz – AL	14 de	14 de	16 d	23 cd	26 cd
Dimilin 3/4oz – AL	8 e	9 e	13 d	18 cd	24 cd
UTC –AL	4 e	6 e	7 d	9 d	9 d
UTC - AN	3 e	4 e	5 d	6 d	7 d

<sup>1</sup> Applied treatment materials to vegetation only. AL = *Aphthona lacertosa*, AN = *Aphthona nigriscutis*.

<sup>2</sup> A one-way analysis of variance with a Tukey multiple comparison procedure determined statistical differences ( $P \leq 0.05$ ). Means in the same column followed by the same letter are not significantly different.

Table 5. Mean percentage mortality of established *Aphthona* flea beetles in treated and untreated field plots from combined locations at three post-treatment intervals.

Treatment	Treated Plots			Untreated Plots		
	1 week	2 week	3 week	1 week	2 week	3 week
Dimilin 1oz	16	47	73	-19	47	63
Dimilin 3/4oz	-39	23	37	-19	47	63
Carbaryl 16oz	83	89	94	-2	45	77
Carbaryl 8oz	55	75	79	-2	45	77
Malathion 8oz	69	85	94	19	61	82
Malathion 4oz	48	71	89	19	61	82
Bran Bait 2lb	54	61	82	19	61	82

Table 6. Mean percentage mortality of *Aphthona* flea beetles in treated and untreated field plots at the three study locations one week after treatment.

Treatment	Study Location		
	Aus	Van Daele	Durham
Dimilin 1oz	-19 a	39 a	28 a
Dimilin 3/4oz	-2 a	-12 a	-101 b
Dimilin untreated	18 a	0 a	-73 b
Carbaryl 16oz	97 a	80 a	71 a
Carbaryl 8oz	53 a	93 a	18 b
Carbaryl untreated	-8 b	24 a	-24 b
Malathion 8oz	92 a	56 a	58 a
Malathion 4oz	86 a	41 a	17 bc
Carbaryl bran bait	32 b	79 a	51 ab
Malathion & bait untreated	54 ab	60 a	1 c

<sup>1</sup> A one-way analysis of variance with a Tukey multiple comparison procedure determined statistical differences ( $P \leq 0.05$ ). Means in the same column followed by the same letter are not significantly different.

Table 7. Mean percentage mortality of *Aphthona* beetles 7 days after exposure to selected grasshopper control treatments in laboratory studies and in field plots.

Treatments	Laboratory Bioassays		Field Plots	
	Insects	Vegetation	Unadjusted	Adjusted <sup>1</sup>
Carbaryl 16	94	96	83	82
Carbaryl 8	95	86	55	60
Malathion 8	41	38	69	44
Malathion 4	32-36	25-67	48	21
Carbaryl Bait	71-90	-	54	17
Dimilin 1	27	26	16	18
Dimilin ¾	12	24	-39	-36
UTC	7-9	7-9	(-19 to 19)	

<sup>1</sup>Connin and Kuitert, 1952.

Table 8. Percentage composition of *Aphthona lacertosa* (AL) and *Aphthona nigriscutis* (AN) at 8 different plots at each of 3 locations.

Treatments	Locations						Mean	
	Durham		Van Daele		Aus		AL	AN
	AL	AN	AL	AN	AL	AN		
Dimilin 1oz	-	-	41	59	100	0	74	26
Dimilin 3/4oz	-	-	0	100	13	87	11	89
Carbaryl 16oz	36	64	0	0	10	90	35	65
Carbaryl 8oz	2	98	24	76	88	12	55	45
Malathion 8oz	91	9	0	100	0	100	79	21
Malathion 16oz	11	89	75	25	21	79	17	83
Bran Bait	54	46	100	0	23	77	54	46
Untreated	10	90	17	83	100	0	50	50
Mean <sup>1</sup>	46	54	24	76	63	37	50	50

<sup>1</sup> Mean percentage based on total counts of all plots and locations.

Table 9. Mean leafy spurge plant density indices for all plots and locations.

	Aus			Van Daele			Durham		
	6/21/00 <sup>1</sup>	6/25/01	7/30/01	6/25/00	6/20/01	7/31/01	6/23/00	6/21/01	8/1/01
Dimilin 1oz	2.6 ab	2.6	2.6	3.0 a	2.9	3.3	3.0 a	1.2	1.0
Dimilin 3/4oz	1.8 c	2.1	2.1	2.7 a	2.6	2.9	2.8 a	2.4	2.8
Carbaryl 16oz	2.7 a	3.1	3.3	2.7 a	2.8	2.8	2.9 a	2.7	2.4
Carbaryl 8oz	2.8 a	2.6	2.4	2.5 a	2.9	3.0	2.8 a	2.8	2.3
Malathion 8oz	2.8 a	3.2	3.1	2.8 a	2.6	2.9	3.2 a	1.2	1.3
Malathion 4oz	2.6 ab	2.8	3.1	2.9 a	2.9	3.3	2.8 a	2.2	2.6
Bran bait	2.6 ab	2.9	3.0	2.5 a	2.7	2.7	2.8 a	1.7	1.1
Untreated	2.0 bc	2.5	2.1	2.9 a	3.0	3.3	2.9 a	2.5	2.1
Mean	2.5			2.8			2.9		

<sup>1</sup> A one-way analysis of variance with a Tukey multiple comparison procedure determined statistical differences ( $P \leq 0.05$ ). Means in the same column followed by the same letter are not significantly different.

Table 10. Grasshopper species composition and age structure for the three study locations.

Species	Test Location – Percent Composition		
	Durham	Van Daele	Aus
<u>Subfamily Gomphocerinae</u>			
<i>Aeropedellus clavatus</i>	-	-	0.61
<i>Ageneotettix deorum</i>	19.66	5.00	11.89
<i>Amphitornus coloradus</i>	1.23	0.45	1.43
<i>Aulocara elliotti</i>	0.25	-	0.20
<i>Aulocara femoratum</i>	1.97	-	-
<i>Chloealtis conspersa</i>	-	0.45	-
<i>Eritettix simplex</i>	0.25	2.27	-
<i>Mermiria bivittata</i>	-	0.45	0.41
<i>Opeia obscura</i>	2.70	10.45	8.61
<i>Orphulella speciosa</i>	3.93	4.09	9.22
<i>Phlibostroma quadrimaculatum</i>	3.19	-	4.10
<i>Pseudopomala brachyptera</i>	-	-	0.20
<u>Subfamily Melanoplinae</u>			
<i>Dactyloptum bicolor</i>	0.25	-	-
<i>Hesperotettix viridis</i>	0.25	-	-
<i>Hypochlora alba</i>	-	0.45	-
<i>Melanoplus bivittatus</i>	4.42	1.36	0.61
<i>Melanoplus dawsonii</i>	0.25	8.18	3.28
<i>Melanoplus femurrubrum</i>	7.37	1.82	7.99
<i>Melanoplus gladstoni</i>	2.21	1.36	0.61
<i>Melanoplus infantilis</i>	0.74	-	7.79
<i>Melanoplus keeleri</i>	1.72	-	2.05
<i>Melanoplus packardii</i>	23.10	0.45	0.82
<i>Melanoplus sanguinipes</i>	16.95	2.73	3.07
<i>Phoetaliotes nebrascensis</i>	5.41	54.55	27.46
<u>Subfamily Oedipodinae</u>			
<i>Arphia pseudonietana</i>	-	-	0.41
<i>Camnula pellucida</i>	0.74	-	3.07
<i>Chortophaga viridifasciata</i>	-	0.91	1.23
<i>Encoptolophus costalis</i>	0.49	4.55	1.43
<i>Metator pardalinus</i>	0.25	0.45	-
<i>Spharagemon equale</i>	0.49	-	-
<i>Trachyrhachys kiowa</i>	2.21	-	3.48
Total no. species	24	18	23
Total mean age	3.70	2.52	2.97

Table 11. Grasshopper species composition and age structure for two common species

Species	Instars					Adult	Total no.	% comp.	Mean instar
	1	2	3	4	5				
<b>Aus</b>									
<i>A. deorum</i>			2	8	12	2	24	11.9	4.6
<i>P. nebrascensis</i>	37	57	26				120	27.5	1.9
<b>Van Daele</b>									
<i>A. deorum</i>				2	7	2	11	5.0	5.0
<i>P. nebrascensis</i>	24	44	51	1			120	54.6	2.2
<b>Durham</b>									
<i>A. deorum</i>		1	4	16	49	8	78	19.7	4.8
<i>P. nebrascensis</i>	2	5	3				10	5.4	2.1



Table 12. Recovery of the *Apthona* flea beetle population in year two at the three study locations.

Treatment	Mean fold increase	Rank Transformation
<b>Aus</b>		
Malathion 8oz	2.79	bc
Malathion 4oz RAATs	2.90	ab
Carbaryl 16oz	5.03	a
Carbaryl 8oz RAATs	1.74	bc
Dimilin 1oz	1.04	d
Dimilin 3/4oz RAATs	2.29	bc
Carbaryl Bait	2.97	ab
Untreated	1.27	cd
Mean of treated plots	2.68	
<b>Van Daele</b>		
Malathion 8oz	1.90	b
Malathion 4oz RAATs	3.11	ab
Carbaryl 16oz	2.71	ab
Carbaryl 8oz RAATs	2.96	ab
Dimilin 1oz	2.39	b
Dimilin 3/4oz RAATs	2.50	b
Carbaryl Bait	9.09	a
Untreated	2.88	b
Mean of treated plots	3.52	
<b>Durham</b>		
Malathion 8oz	1.34	c
Malathion 4oz RAATs	3.82	ab
Carbaryl 16oz	5.22	a
Carbaryl 8oz RAATs	3.78	a
Dimilin 1oz	2.08	c
Dimilin 3/4oz RAATs	3.39	ab
Carbaryl Bait	1.43	c
Untreated	2.10	bc
Mean of treated plots	3.00	

<sup>1</sup> A one-way analysis of variance with a Tukey multiple comparison procedure determined statistical differences ( $P \leq 0.05$ ). Means in a column within a study location followed by the same letter are not significantly different.

Table 13. A comparison of year one and year two leafy spurge plant density estimates.

Treatment	Percentage of initial plant density estimate – June 21-25, 2000 <sup>1</sup>						Mean % change
	6/20 – 6/25/01			7/30 – 8/1/01			
	Aus	Van Daele	Durham	Aus	Van Daele	Durham	
Dimilin 1oz	102 ab	97 a	40 c	103 ab	110 a	34 c	-18
Dimilin 3/4oz	125 ab	95 a	86 ab	119 ab	110 a	98 a	+9
Carbaryl 16oz	119 ab	103 a	98 ab	121 a	105 a	83 a	+3
Carbaryl 8oz	91 b	118 a	103 a	87 b	123 a	87 a	-1
Malathion 8oz	113 ab	97 a	38 c	111 ab	111 a	41 bc	-12
Malathion 4oz	107 ab	100 a	79 ab	121 a	112 a	94 a	+9
Bran bait	111 ab	110 a	65 bc	114 ab	110 a	40 c	-12
Untreated	132 a	105 a	87 ab	113 ab	115 a	71 ab	0

<sup>1</sup>Data evaluated by a Kruskal-Wallis one way analysis of variance on ranks. Dunn's method (an all pairwise multiple comparison procedure) determined differences between treatments (P<0.05)

Figure 1. Tower-mounted airbrush (Paasche Type H with R75 regulator) used in simulating aerial applied sprays in laboratory studies.

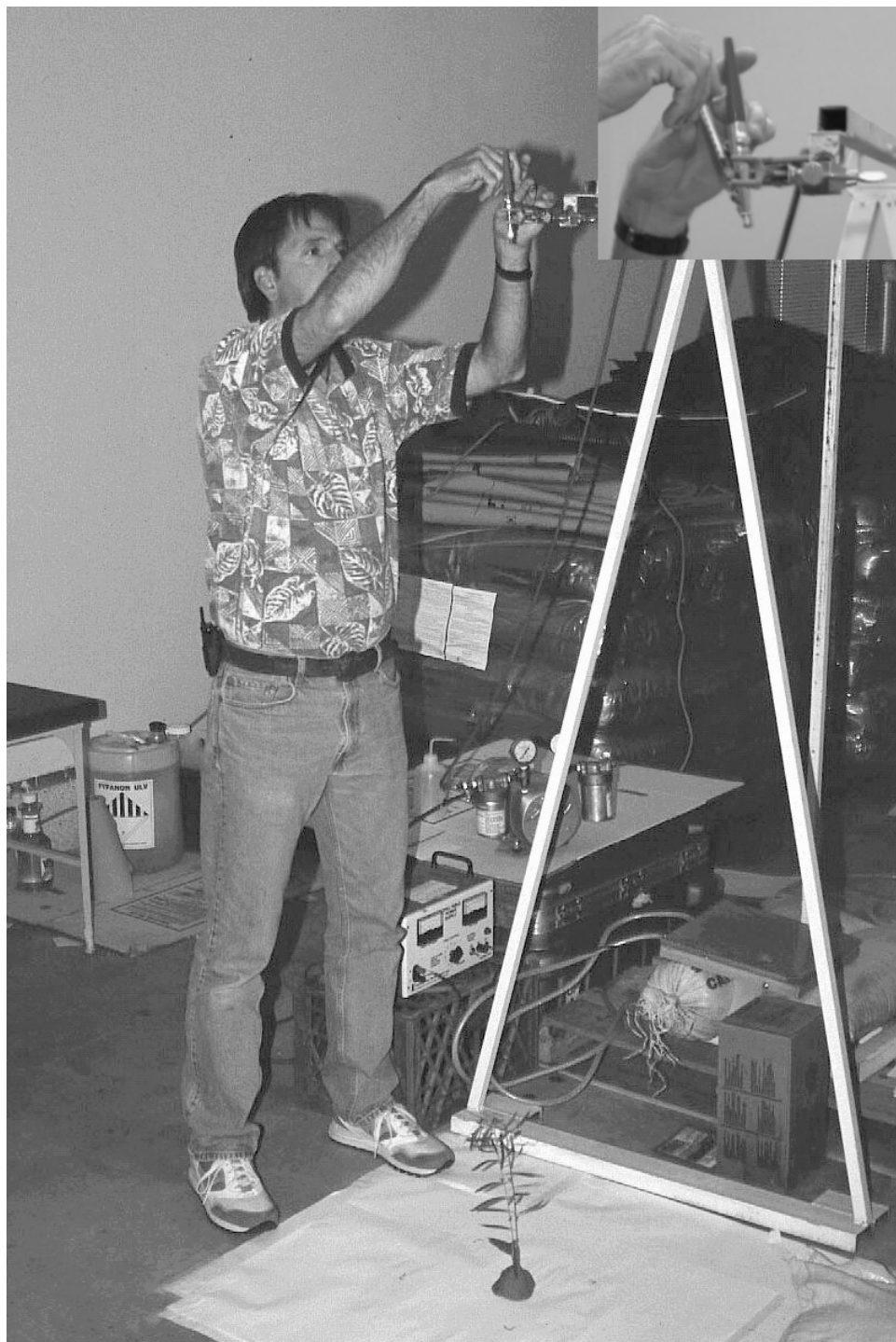


Figure 2. Location of the three study sites in western North Dakota.

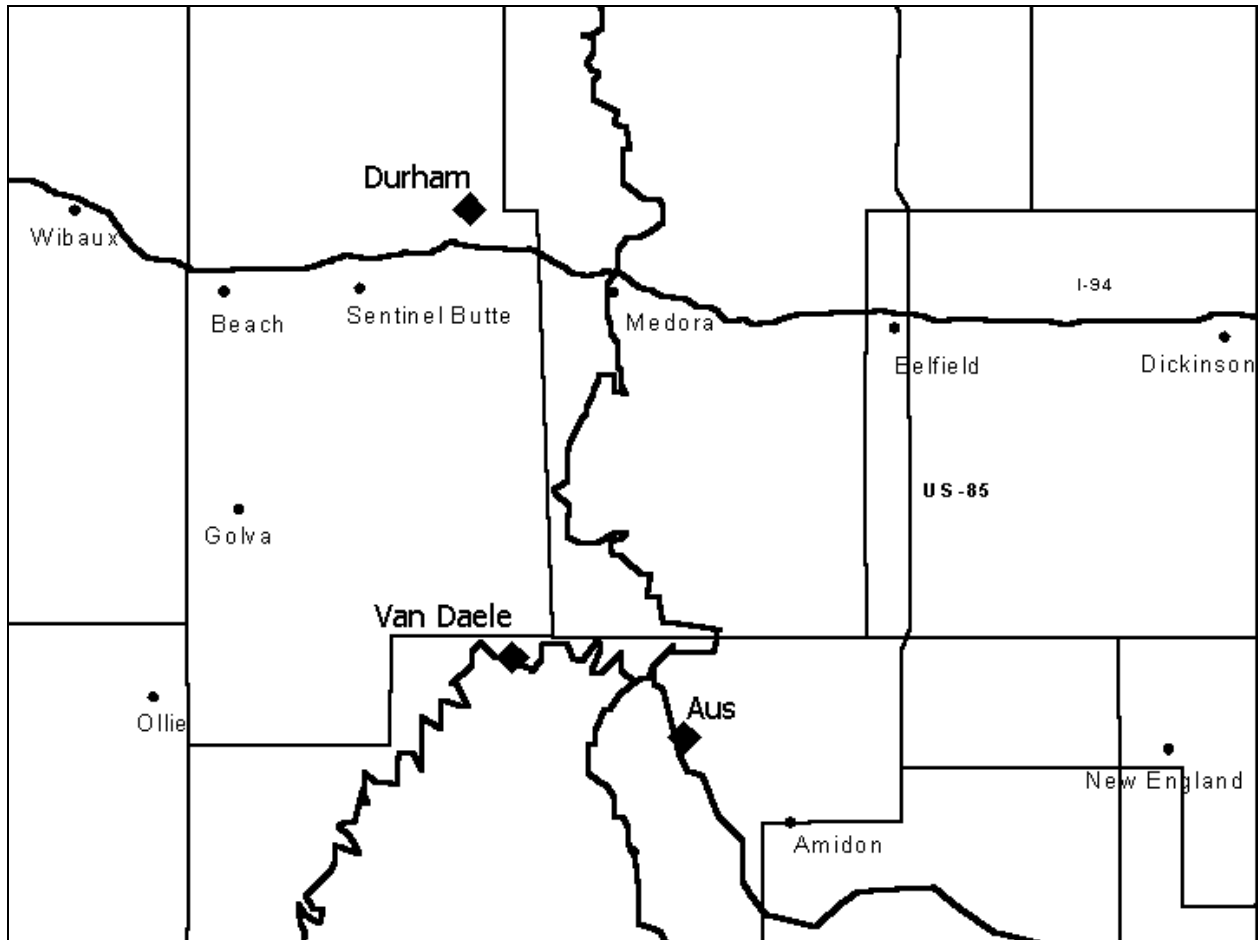


Figure 3. Locations of the 8 research plots (as determined by GPS data) at the Aus ranch study site.

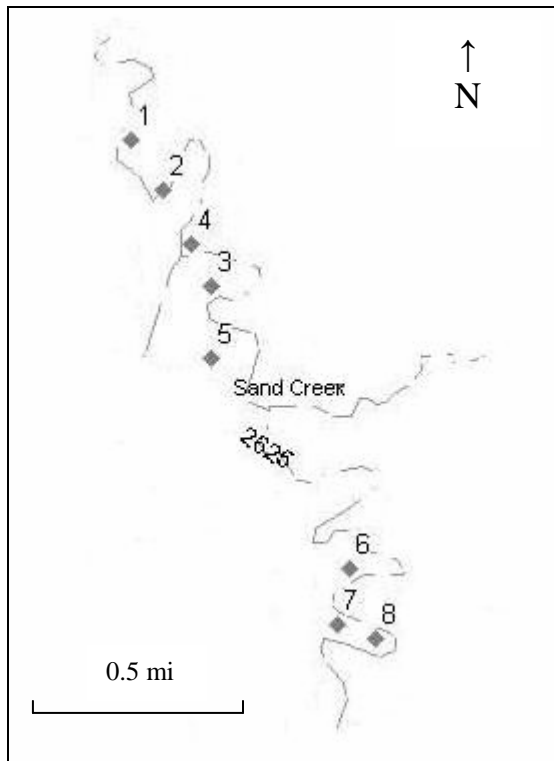


Figure 4. Locations of the 8 research plots (as determined by GPS data) at the Van Daele ranch study site.

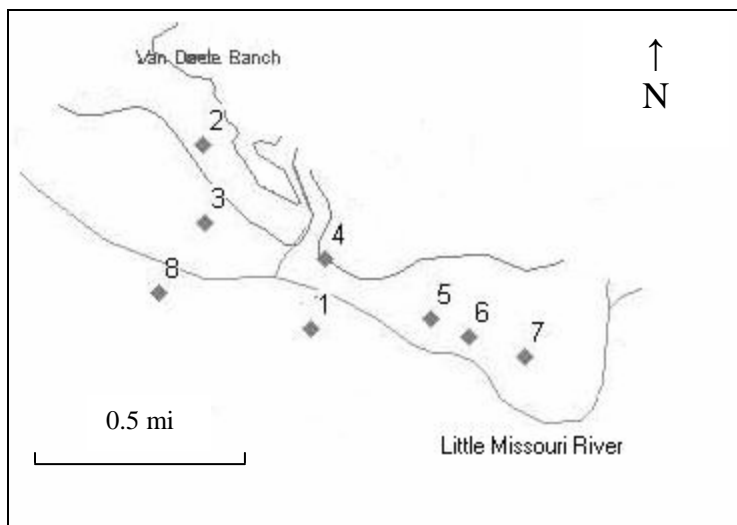


Figure 5. Locations of the 8 research plots (as determined by GPS data) at the Durham ranch study site.

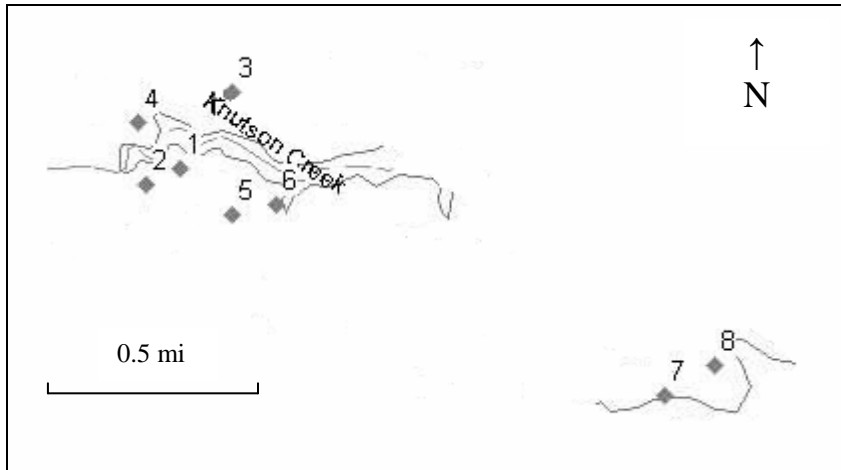


Figure 6. Plot diagram of 16 sampling sites, spray card locations and application direction.

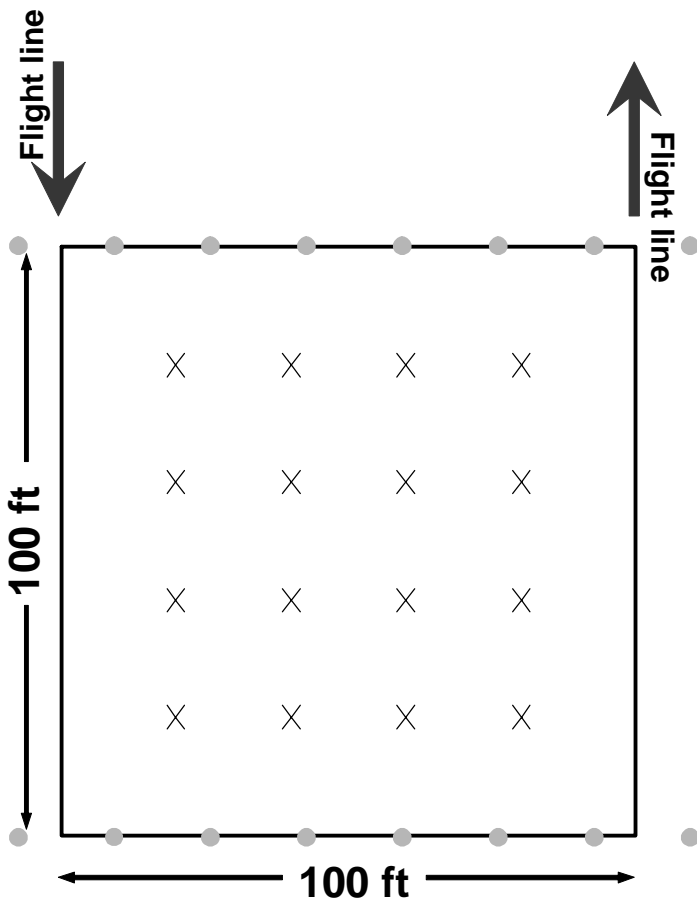


Figure 7. Accumulated daily precipitation records for July, 2000 for all three locations.

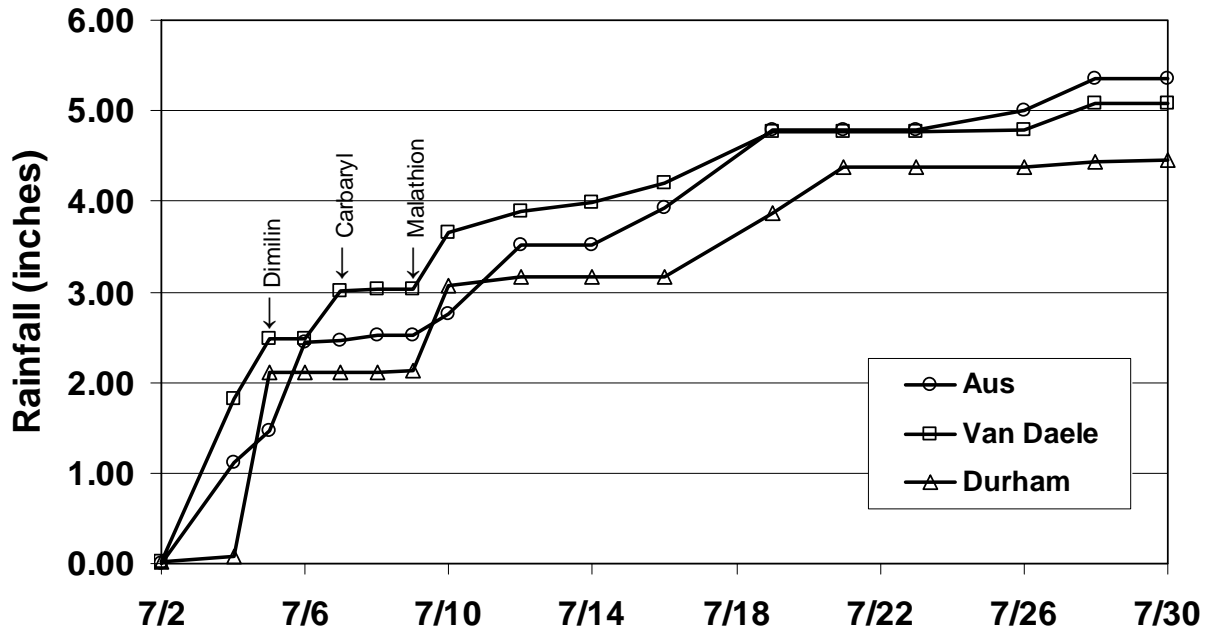


Figure 8. Examples of leafy spurge index values: (A) – no leafy spurge plants in the sweep area = 1, (B) – one to a few plants in the sweep area = 2, (C) – many plants but not continuous coverage in the sweep area = 3 and (D) – continuous coverage of plants in the sweep area = 4.





Figure 9. Adult *Aphthona* densities in the untreated control populations at the three study locations in 2000.

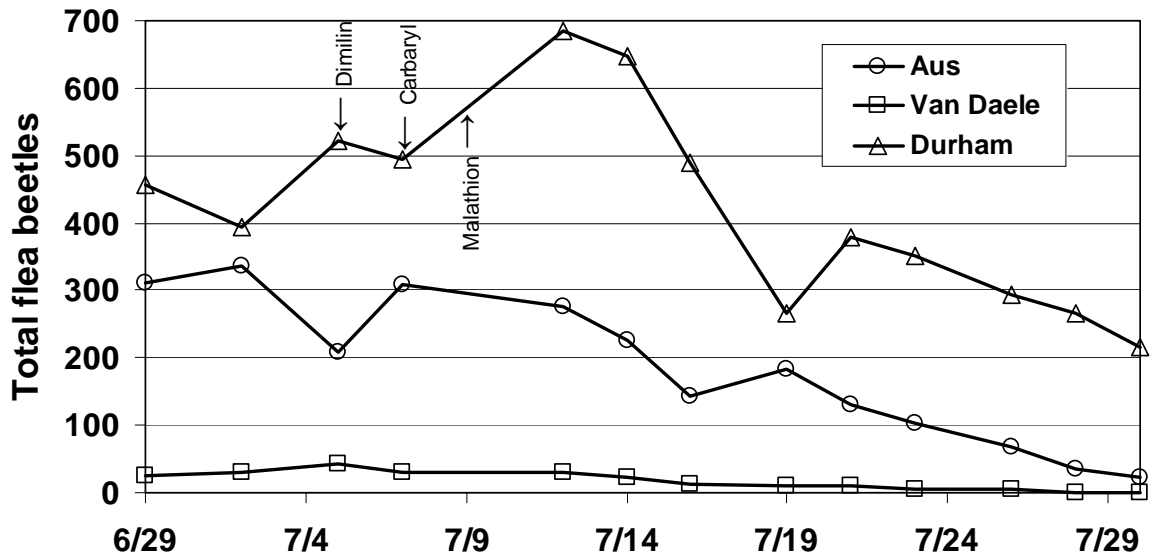


Figure 10. Adult *Aphthona* densities in the untreated control populations at the three study locations in 2001.

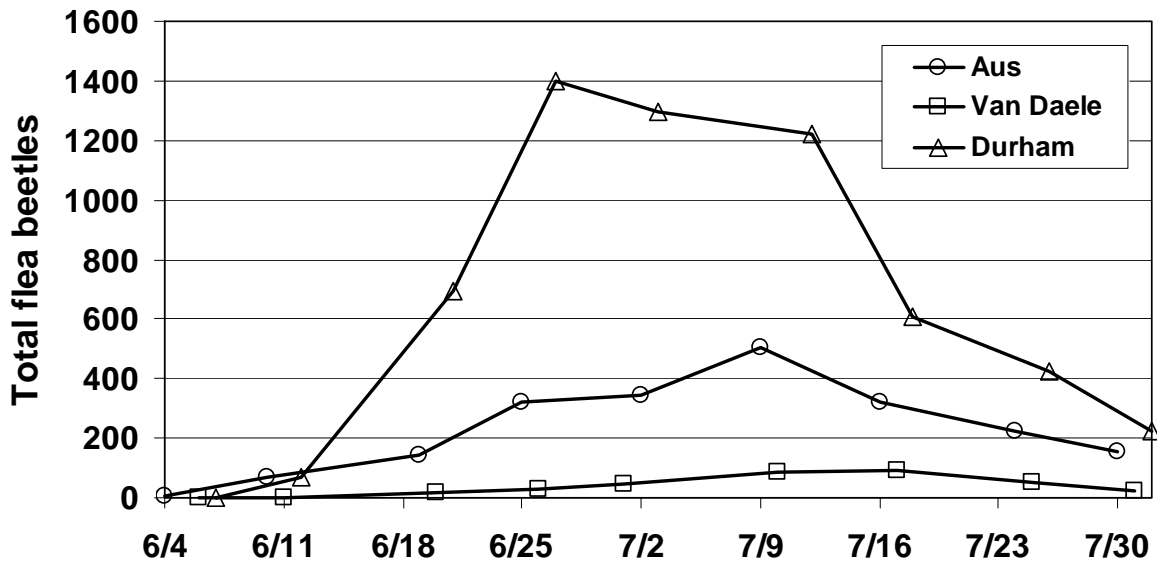


Figure 11. Comparison of 2000 and 2001 *Aphthona* population densities at the Aus study location.

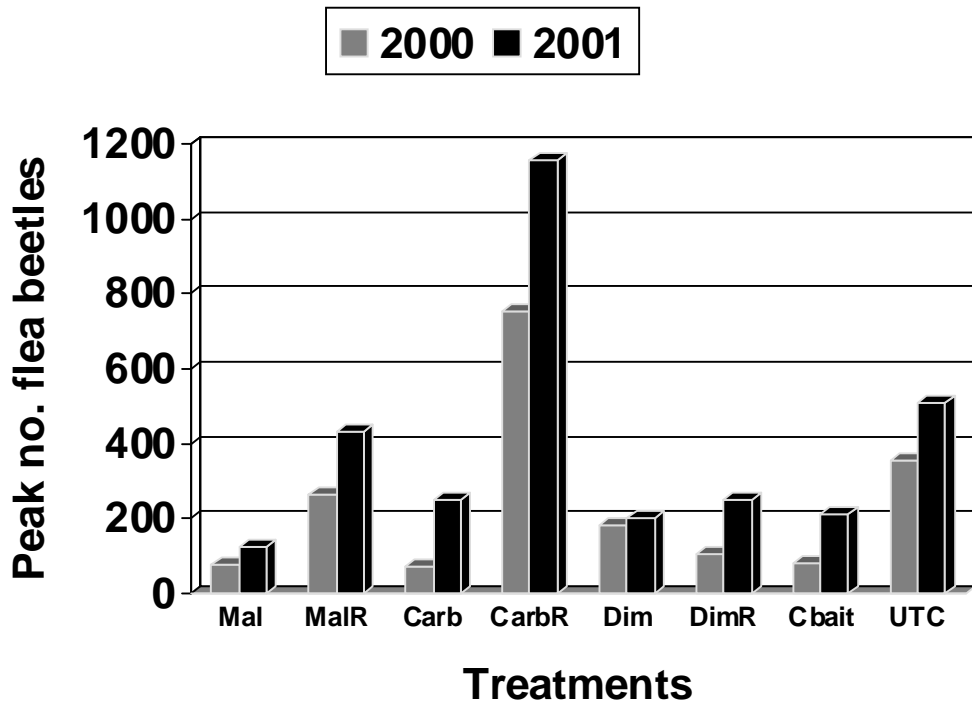


Figure 12. Comparison of 2000 and 2001 *Aphthona* population densities at the Van Daele study location.

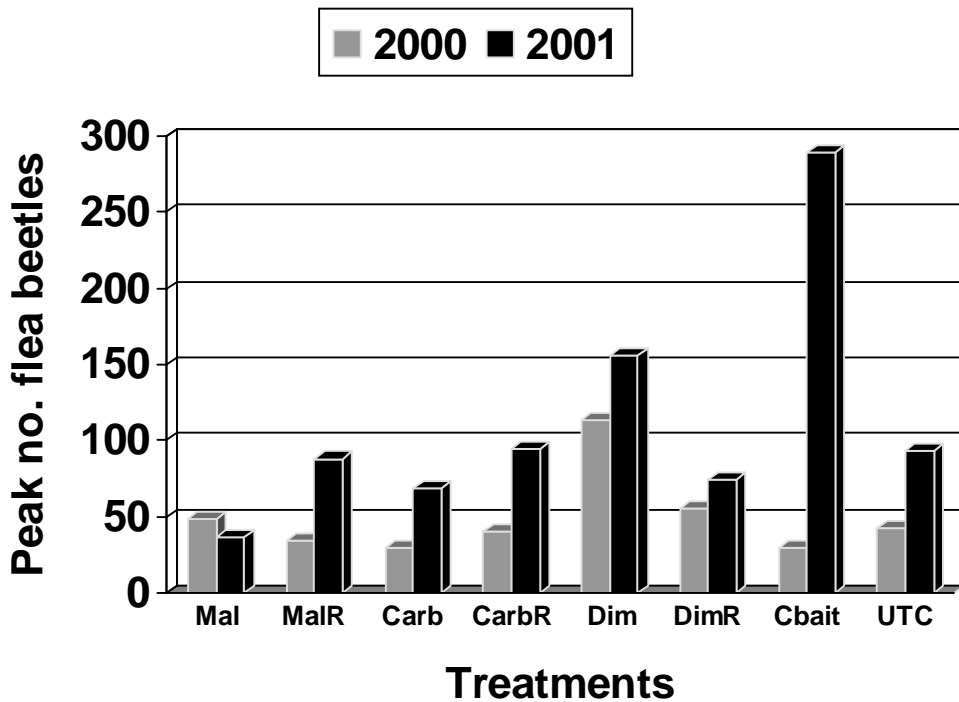


Figure 13. Comparison of 2000 and 2001 *Aphthona* population densities at the Durham study location.

